Contributions from the Cryptogamic Laboratory of Harvard University. XXIII.[1]

Notes on the life history of a blue-green motile cell.

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WITH PLATE XI.

In view of certain opinions that have been expressed by some botanists, notably Hansgirg, but which most botanists have not generally accepted, that there exist motile conditions of certain members of the MYXOPHYCEÆ (CYANOPHYCEÆ), the writer was much pleased when last November he happened to meet with a unicellular blue-green motile organism and was able to trace its life history.

These blue-green motile cells were first noticed while examining some material collected from a pool in the salt marshes of the Charles river, Cambridge. They appeared in such quantities in vessels holding the collections of *Beggiatoa*, *Oscillatoria*, *Melosira*, *Cladophora*, *Enteromorpha clathrata*, etc., found in the salt marshes, that they formed a scum upon the surface of the water and sides of the vessels, where they readily passed into a non-motile stage. The source of the blue-green swarmers was soon traced to colonies of bluish-green cells, that resembled colonies of Polycystis, and were found commonly adhering on the sides of marsh grass (Spartina) and other objects. The measurements of these cells were identical with those of *Polycystis pallida*, and the colonies resembled strikingly the herbarium specimens of this species.

Anton Hansgirg has been the most prominent champion of polymorphism among the Myxophyceæ. He has expressed himself as believing that species of Euglena[2] give rise to Oscillatoria filaments and certain blue-green swarmers he thinks may come from Oscillatoria filaments[3] although he has not seen their formation. Goebel[4] states that he has seen the formation of swarm spores in Merismopedia. Thus it was

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[1] Prepared under the direction of Dr. W. G. Farlow.

[2] Hansgirg, Botanisches Centralblatt **23:** 232. 1885.

[3] Hansgirg, ibid. p. 230.

[4] Goebel, Outlines of Class. and Sp. Morph. of Plants (Engl. trans.) 22. 1887.

with considerable interest that the writer undertook the study of the form which he had found whose non-motile state so closely resembled *Polycystis pallida.*

Motile stage.

The motile cells were to be found at all times in small numbers. There was no time of day when they appeared in quantities, as is the habit of zoospores of members of the Chlorophyceæ, although they exhibited the same phenomena of collecting on the sides of the vessel nearest the light. When confined in a Van Tieghem cell they swarm about for a day or two, finally coming to rest at the edge of the drop of water.

The cells (plate XI, fig. 1) are broadly elliptical in outline, from 8–10μ long and 5–6μ wide. One end is slighly truncate in shape and contains a slight depression into which the pair of cilia are inserted. The cilia are not the same length, the longer being about as long as the cell is wide and the other somewhat shorter. They are so placed, and the figure illustrates this point, that the longest cilium is nearest to the longest axis of the cell.

Inside the cell are from six to ten disc-shaped bodies arranged around the periphery of the cell. These bodies are not pyrenoids, nor are they amorphous albuminous matter, for they are readily destroyed by a dilute potassic hydrate solution after long treatment in mercuric chloride in absolute alcohol. For this reason it seems as though these bodies are true chromatophores, although the blue-green tint is not always confined to them. Sometimes the blue-green color seems to fill almost the entire cell, only the end which bears the cilia being hyaline.

Near the middle of the cell on the periphery are one or often two bright red pigment spots: when two they are always placed near together, sometimes almost touching. The presence of the two pigment spots in one individual did not seem to indicate that conjugation had taken place, for such specimens were not necessarily larger in size and no specimen was ever observed with four cilia.

At the opposite end of the cell from the cilia there is usually to be found a light colored area that probably marks the position of the nucleus, which was demonstrated to be present in the non-motile cells.

The motion of the motile cells through the water is quick and sometimes they dart to one side in a manner that suggests at once the motion of certain infusoria. They come to rest slowly, moving occasionally from side to side some time after they seem to have settled down. While they sometimes spin around on the ciliated end just before they settle down, they do not attach themselves in a perpendicular position but rest on one side. The length of time these cells remain in the motile condition, and the character of their movements, forms a striking contrast to the motile phases of other algæ, such as Cladophora, Draparnaldia, Ulva, etc., whose zoospores settle down within a few hours to develop a new individual.

Non-motile stage.

The motile cells when they came to rest in a Van Tieghem cell, did not divide for two or three days and after that only at intervals of two or three days. The division consists of a longitudinal splitting of the cell into a pair of similar cells and hence in the colonies they tend to show a pretty regular arrangement in groups of twos and fours. They are about the same size as the motile cells but inclined to be a little shorter and somewhat broader in their proportions. That there is a common gelatinous envelope surrounding young colonies is often clearly shown by the quantities of bacteria that swarm at a fixed distance from the cells (plate XI, fig. 2). This gelatinous envelope is not a marked character however and in large colonies it is quite insignificant (plate XI, fig. 3), although it is not difficult to prove its existence with proper stains.

The chromatophores, in the non-motile cells, are not arranged in any regular manner and sometimes the entire cell appears of an almost uniform blue-green tint. The chromatophores vary in size but are usually discoid in shape, (plate XI, fig. 4). They may be brought out with great distinctness in cells after treatment with absolute alcohol, and appear perfectly homogeneous.

One or two, and rarely three, pigment spots are to be found near the middle of the cells on the periphery. Their color however is not so bright as in the motile stage but inclined to be a brownish-red.

A well defined nucleus (plate XI, fig. 5) can easily be demonstrated when specimens, well fixed and hardened, are

treated with dilute potassic hydrate solution to destroy the chromatophores and then stained with eosin.

In several instances, while examining under the microscope the colonies scraped from the marsh grass, single cells were observed to slip out from the colony and swim away and these were identical with the blue-green motile cells.

Taxonomy.

The presence of a well defined nucleus, pigment spots and chromatophores clearly showed that this form was not a member of the Myxophyceæ as the group is defined by most botanists.

Of late there has been considerable discussion on the inner structure of the cells of Myxophyceæ, particularly in respect to the possible occurrence of nuclei and chromatophores. Almost all the investigators in this field of research agree in saying that there never exists a nucleus in the usual sense of the word; that, if present at all, it is in the form of some central body of very indefinite outline, or scattered through the cell in the shape of small granules which react with stains in a manner similar to the chromatin bodies in the nuclei of higher plants. The nucleus in the present form is perfectly well defined and contains a distinct nucleolus. In regard to the presence of chromatophores in the cells of Myxophyceæ there is less unanimity of opinion among recent writers. Several investigators, Hieronymus, Zukal, and others, believe that chromatophores exist; but other investigators, Zacharias in particular, have not been able to find them. The chromatophore of Hieronymus is present as a network just under the cell wall and is only made apparent by special treatment. No investigator appears to have seen bodies as definitely organized and outlined as the chromatophores in the cells of the form that has just been described. For a general review of the literature on this subject the reader is referred to a paper by Flahault in the Revue gén. de Botanique **5:** 181. 1893.

Blue-green motile cells have been known for a long time. Ehrenberg[5] in 1838 published an account of three species under the genera *Cryptomonas* and *Cryptoglena* and since then very little has been added to our systematic knowledge of these forms. The genus *Cryptoglena* was distinguished from *Cryptomonas* by Ehrenberg because of its pigment spot, and

[5]Ehrenberg, Die Infusionsthierchen als vollkommene Organismen. 1838.

therefore the present form clearly belongs to *Cryptoglena*, as defined by Ehrenberg, but no species of Ehrenberg agrees with it in measurement nor does it resemble any of Ehrenberg's figures. Moreover, none of Ehrenberg's species were marine.

Under the name *Cryptomonas polymorpha*, Perty[6] has included all the blue-green forms of Ehrenberg as well as some grass-green forms, but his uniting so many various forms under one name does not seem to be warranted and has not been generally accepted by recent writers. Stein[7] gives an account of the structure of a form he calls *Cryptomonas ovata* Ehrenb.; but, as Hansgirg[8] has shown, his form cannot be the same as Ehrenberg's *Cryptomonas ovata* for the latter is grass-green and has only one cilium, while Stein's form is blue-green and has two cilia. Cienkowsky,[9] previous to Stein's publication, had minutely described a form, calling it *Cryptomonas ovata*, which is evidently much the same form as Stein's *Cryptomonas ovata*. More recently Dangeard[10] has given a description of *Cryptomonas ovata* and his seems also to be the same as that of Cienkowsky and Stein. This form, studied by Dangeard, Stein and Cienkowsky, is very different from that which I have found. Their species has no pigment spot, no bodies corresponding to the chromatophores in my form, it is much larger and the cell shows a degree of complication far greater than is found in the one here described.

Dangeard[11] has also described a new blue-green motile cell, *Cryptomonas cyana*, which is very small (3–4μ long), and quite recently[12] he mentions having observed a marine *Cryptomonas* (*C. marina*), but the description is too short to enable me to judge whether or not it is similar to the present organism. A species that closely resembles my form in measurements, has been described by Hansgirg[13] as *Chroomonas Nordstedtii*, but this species differs in having no pigment spots and in the shape of the chromatophore which is lamina-like with pyrenoids 3μ wide.

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[6] Perty, Zur Kennt. kleinster Lebensf. nach Bau. Funk. System. etc., 1852.

[7] Stein, Der Organismus der Infusionthiere. III Abtheilung, I Hälfte. *pl. 19. fig. 26.* 1878.

[8] Hansgirg, Bot. Centralblatt. **24:** 342. 1885.

[9] Cienkowsky, Ueber Palmellaceen und einige Flagellaten. Archiv f. mikr. Anat. **6:** 424. *pl. 23.* 1870.

[10] Dangeard, Contribution a l'étude des organismes inférieurs. Le Botaniste II.—:47. 1890.

[11] Dangeard, l. c., p. 54.

[12] Dangeard, Note sur un Cryptomonas marin. Le Botaniste III.—:—1892.

[13] Hansgirg, Bot. Centralblatt **23:** 230. 1885.—**24:** 376. 1885.

As the presence of pigment spots, character of the chromatophores and general simplicity of the cell distinguish the form here described from the genus Cryptomonas as understood by Dangeard, Stein, Cienkowsky, and primarily by Ehrenberg, it has seemed best to adopt the name Cryptoglena with the character of the genus as defined by Ehrenberg, and as no described species appears to be like the present form, it seems desirable to publish its description as:

Cryptoglena Americana, sp. nov.—Motile cells broadly elliptical, 8–10μ long, 5–6μ wide: cell contents blue-green with 6–10 disc-shaped chromatophores arranged around the periphery: one or two bright red pigment spots placed on the periphery near the middle of the cell: one end hyaline, slightly truncate, with a depression from which arise a pair of cilia of unequal length, the longer about as long as the cell is wide.

Non-motile cells slightly shorter and somewhat broader than motile cells (7–9μ long × 6–7μ wide), arranged in groups of twos and fours in a closely packed Polycystis-like colony: almost uniformly colored, blue-green, with 6–10 disc-shaped chromatophores and one or two brownish red pigment spots near the middle of the cell at the periphery: nucleus near the middle of the cell.

Habitat: salt marshes of the Charles river, Cambridge, Mass., on stems of grass and larger algæ. Autumn.[14]

The agreement in measurements between the non-motile cells of *Cryptoglena Americana* and the cells of the American form of *Polycystis pallida*, described by Dr. Farlow,[15] is very interesting and suggests the possibility that this *Polycystis pallida* is really the non-motile condition of this species of Cryptoglena. I have had the opportunity of comparing herbarium specimens of the American *Polycystis pallida* with European specimens of the same species and they seem to be identical.

Herbarium speeimens of *Polycystis pallida* give very little evidence of a cell structure as differentiated as *Cryptoglena Americana*, but specimens of *Cryptoglena Americana* after being dried on mica for a month gave very little indication of the chromatophores and no indication of the pigment spots.

[14]The motile condition appears to be common all through the winter. It has frequently been met with since the above was written.

[15]The marine algæ of New England 28, 1879.

The question of a possible identity of these two forms can only be settled by an examination of fresh material of *Polycystis pallida* and it is desirable that botanists who have the opportunity of investigating this point should bear the question in mind.

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EXPLANATION OF PLATE XI.

All figures sketched with an Abbé camera and magnified about 750 diameters; reduced in engraving one-tenth.

Fig. 1, Motile cells, killed with Flemming's fluid.—Fig. 2, A young colony of non-motile cells; the boundary of a surrounding gelatinous envelope is marked by the swarm of bacteria.—Fig. 3, A colony of non-motile cells.—Fig. 4, Four non-motile cells after treatment with absolute alcohol, showing chromatophores.—Fig. 5, Two non-motile cells, after treatment with dilute potassic hydrate solution, and staining with eosin, showing nuclei.